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EFFECT OF AERIAL EXPOSURE ON PHYSIOLOGICAL CONDITION AND SURVIVAL OF *DIPLODON CHILENSIS* (BIVALVIA: HYRIIDAE) DURING TRANSLOCATION

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INTRODUCTION

Freshwater mussels are an important component of the aquatic biodiversity of Chile, especially in the southern part of the country, and other geographical areas of the northern and southern hemispheres. The most common species in Chile is *Diplodon chilensis* (Gray, 1828), which is a unique species of Hyriidae with a distribution extending from 34°58'S to 72°48'S in both lentic and lotic environments in numerous hydrographic basins (Parada & Peredo, 2002). It is also present in Argentina between parallels 32°52' and 45°51'S (Bonetto, 1973). *Diplodon chilensis* populations have declined or been extirpated in lotic environments due to disturbance or habitat degradation from anthropogenic activity, mainly engineering projects. To date there have been no proposals in Chile for the protection of freshwater mussels. Relocation of unionids is being used as a conservation and management tool (Cope & Waller, 1995). They reviewed 33 projects and reported a mean survival of 50%, suggesting that relocation had been stressful to mussels. Furthermore, most relocations were monitored for < 1 year, and < 20% were monitored for five years. The success of most relocation projects has been predominately judged by mussel survival. Few studies examined measures such as growth, recruitment or condition index.

Peredo et al. (2005), studying relocation of *D. chilensis* in Chile and its long-term evaluation, reported positive results due to recruitment and recovery of mussels 18 years after relocation. However, little is known of the effect of handling and aerial exposure on mussel survival or recommendations on the most appropriate season to conduct relocations. It is hypothesized that relocation of freshwater mussel populations is a suitable management and conservation strategy if the stress of handling is minimized and time of the year fits with the physiological condition of mussels, the period when reproductive stress is low and metabolic rate sufficient for reburrowing into substratum. The goal of the present study was to evaluate the effect of aerial exposure on survival and physiological condition of *Diplodon chilensis* at two different times of the year, through short-term relocation experiments.

MATERIAL AND METHODS

Study Site

Lake Colico is located at 39°07'S, 72°00'W, in the pre-Andes Mountains region. The lake is part of the Chilean northern-patagonian lakes called the Araucanian lakes and belonging to the Toltén River basin. It is pristine, oligotrophic and with low species diversity. In summer, its temperature ranges between 19.5°C and 21°C and in winter, between 9.2°C and 9.4°C. Its maximum depth is 250 m.

The fish farm channel receives water from the Curaco River, an outlet of Lake Colico. Mean water temperatures at the fish farm of the School of Aquaculture of the Catholic University of Temuco at Los Laureles (39°2'S, 72°12'W) range between 9.6°C in August (winter) and 18.9°C in February (summer).

In late fall-winter and spring-summer periods of 2003, *D. chilensis* were randomly sampled by scuba diving at 2 m depth from Lake Colico and transported during 20 min to the fish farm in aerated coolers of 40 x 25 x 35 cm with lake water, located at Los Laureles 30 km from Lake Colico. The mussels were placed in the fish farm channel, where the experiments were conducted.

The experiment design of fall-winter and spring-summer studies consisted of three aerial exposure treatments (2, 4, 8 h), with three replicates per treatment and respective controls,

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TABLE 1. Mean water temperature, relative humidity, air temperature, and air temperature changes during exposure treatments with mussels in winter and summer study periods.

Study Period	Lake T in °C	Channel T in °C	Exposure duration in hours	Relative Humidity in %	Air T initial in °C	Air T end in °C	Δ T in °C
Winter	10.1	9.8	2	91	6.5	7.3	0.8
			4	91	6.5	8.0	1.5
			8	91	6.5	12.5	6.0
Summer	14.0	14.0	2	73	12.5	17.0	4.5
			4	73	12.5	17.6	5.1
			8	70	12.5	22.1	9.6

which were not emersed. Each treatment and its respective control for fall-winter and spring-summer periods consisted of 75 mussels (25 per replicate), chosen at random from the total mussels collected, and placed in wire cages (30 x 20 x15 cm mesh) to prevent escapement and predation. The bottom of each cage was covered with the channel substratum, and the cages were marked for later identification.

Prior to placing mussels into the cages, individual valve lengths (mm, anterior to posterior) and wet weights (g) were measured to determine condition index, using the relation CI = WTM/L^3 x 100 (WTM = wet total mass; L = valve length) adapted from Newton et al. (2001). Wet total mass corresponds to the total weight of the specimen (valve weight plus soft tissue weight). We used wet mass to avoid sacrifice of animals. Air temperature and relative humidity were recorded during the time of aerial exposure of the mussels.

Three months after relocation, mussels were examined for survival (live vs. dead) and physiological condition. Two-way ANCOVA was used to establish statistically significant differences between initial condition index (iCI) and final condition index (fCI) of each of the treatments and respective controls, between final condition index (fCI) of treatments and controls, and between final condition index (fCI) of treatments of winter and summer studies. Differences among treatments were judged significantly at $P < 0.05$ level.

RESULTS

Aerial Exposure Period

All mussels subjected to 2, 4, and 8 h of aerial exposure and the respective controls in the fall-winter and spring-summer periods survived the initial treatments. However, some mussels of all treatments showed signs of stress such as gaped valves and protruded feet. Evidence of stress in mussels was observed in experiments conducted in both periods of the year, with stress signs becoming more evident in mussels exposed for 8 h.

Water temperature of Lake Colico and that of the fish farm channel did not show significant differences in the experiments carried out concurrently. However, differences were observed between experiments performed at different periods of the year. Thus, lake water and channel water temperature in spring-summer period was 4.2°C higher than in fall-winter period (Table 1). Air temperature during the 8 h exposure treatment was higher in spring-summer period (9.6°C) than in fall-winter period (6.0°C) (Table 1).

Resurvey Period

After three months in the channel, mussels did not showed differences in survival between experiments carried out in the fall-winter period. Mussels did showed differences in treatments conducted in the spring-summer period (Fig. 1). In fall-winter period, survival was

TABLE 2. Values of p ANCOVA comparing initial Condition Index (iCI) and final Condition Index (fCI) of treatments and controls of winter and summer studies (p-value < 0.05).

fCI vs iCI	Winter p value	Summer p value
control	0.526	0.345
2h	0.13	0.984
4h	0.897	0.506
8h	0.841	0.764

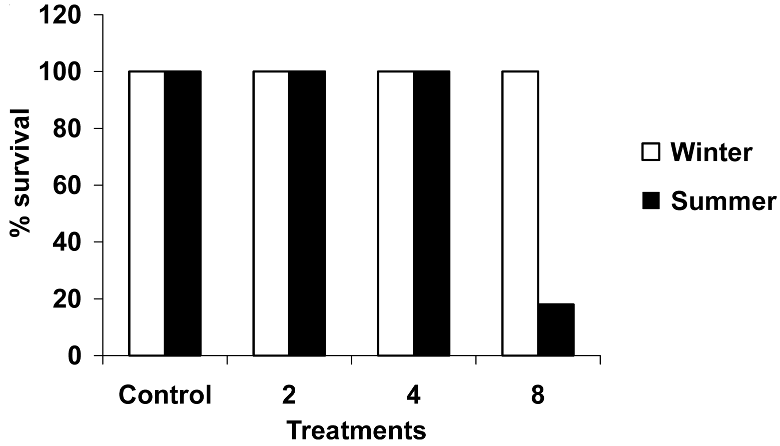

FIG. 1. Percent survival of 0 (control), 2, 4, and 8 hours emersion treatment groups of mussels of summer and winter study.

100% in each of the treatments (2, 4, and 8 h of air exposure) and the respective controls. In the spring-summer period, the 2 and 4 h aerial exposure treatments, survival was 100%, and 18% in 8 h air exposure treatment (Fig. 1). The control survival was 100%. In two of the replicates of the 8 h treatment, survival was 0% (survival was not observed in 100% of the mussels).

Two-way ANCOVA showed no significant differences between initial condition index (iCI) and final condition index (fCI) of each of the treatments and respective control (Fig. 2, Table 2), nor were there significant differences between final condition index (fCI) of treatments and controls, as well as between final condition index (fCI) of treatments of winter and summer studies (Tables 3, 4). In the spring-summer test we did not consider two replicas from 8 h treatment because the mussels were dead.

DISCUSSION

The differences in survival observed in the winter experiment relative to the summer experiment is attributable to air temperature and relative humidity rather than to the duration of aerial exposure. The former factors contribute to dessication of mussels. The locality where the experiments were conducted has a mediterranean climate, determined by the high relative humidity present from April to November (Di Castri & Hajek, 1976). Studies carried out by Dietz (1974) Byrne & McMahon (1994), Waller et al. (1995), and Bartsch et al. (2000) corroborate our results.

Diplodon chilensis is a tachytictic species, being reproductively active during spring-summer, completing glochidial development in late summer (Peredo & Parada, 1986). These features could explain the survival differences in winter vs. summer. In summer, the mussels

TABLE 3. Values of p ANCOVA comparing control and each of the treatment final Condition Index (fCI) of winter and summer studies (p-value < 0.05).

Treatment and respective controls fCI	Winter p value	Summer p value
2 h	0.492	0.871
4 h	0.541	0.602
8 h	0.568	0.506

TABLE 4. Comparison among treatment final Condition Index (fCI) of winter and summer studies (p-value < 0.05).

Treatment fCI	Winter p value	Summer p value
ICf 2h v/s ICf 4h	0.9	0.465
ICf 2h v/s ICf 8h	0.093	0.102
ICf 4h v/s ICf 8h	0.102	0.967

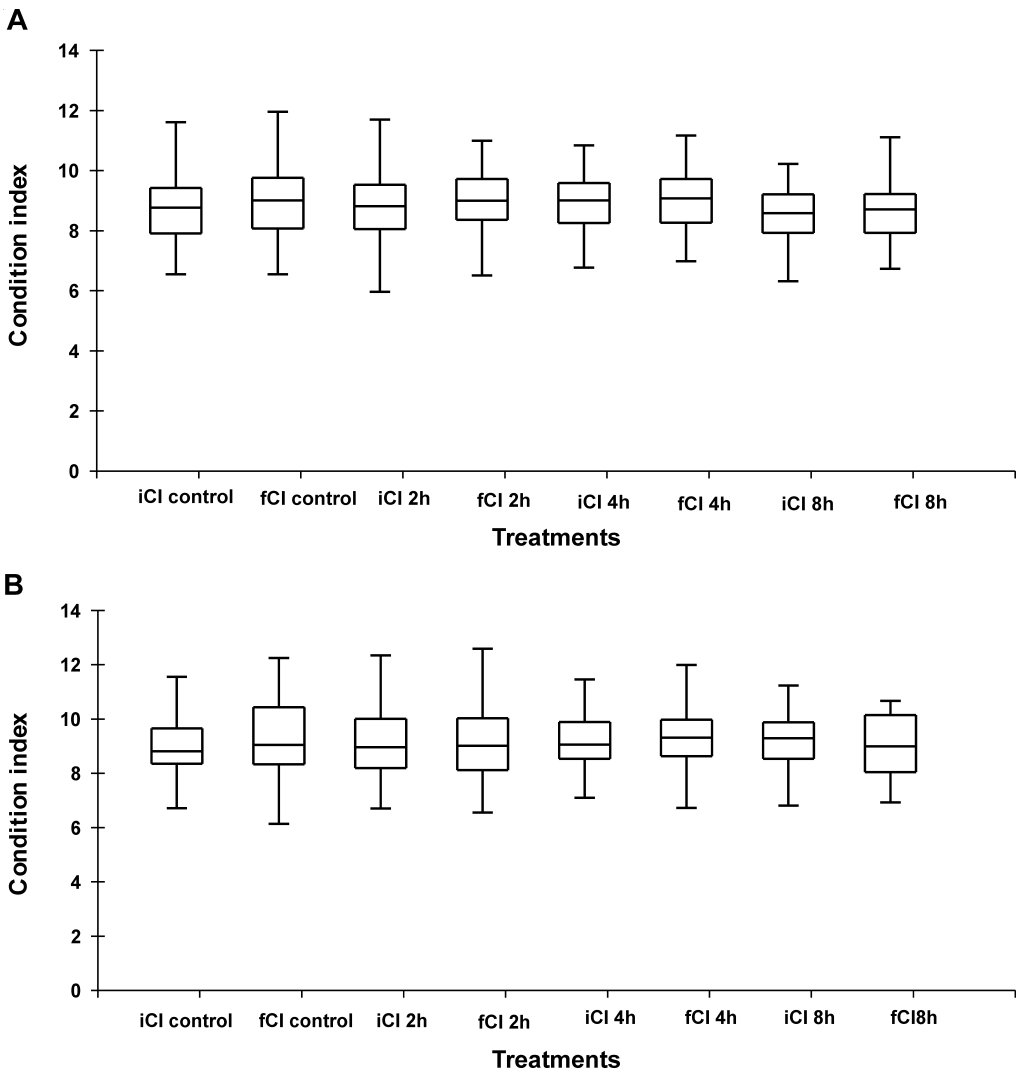

FIG. 2. Mean initial Condition Index (iCI) and final Condition Index (fCI) during 3 months of relocation into channel of mussels exposed to 0 (control), 2, 4, and 8 hour treatment (A = winter study; B = summer study).

would be more vulnerable to perturbation (aerial exposure) due to their reproductive condition, as was seen in individuals with gaping valves when exposed for 8 h and temperatures above 20°C. Conversely, winter low temperatures and high relative humidity would allow individuals to tolerate longer exposure periods. Similar results have been reported by Waller et al. (1995) in fall and spring for *Amblema plicata,* a tachytictic species.

With regard to the physiological condition of *D. chilensis* after three months in the fish farm channel, results show no significant differences between values of the CI before and after the aerial exposure, in survivors of the three treatments at the same time of year, neither between experiments conducted in winter versus summer. Specimens that survived the treatments retained their initial condition after relocation at any time of the year.

Our results coincide with those of Newton et al. (2001) for mussels relocated to a river.

In conclusion, we found that air temperature and relative humidity during emersion are the factors that most effect mussel survival and that after three months, and surviving mussels exhibited a physiological condition similar to that recorded before relocation. The most appropriate time to relocate *D. chilensis* populations is the fall-winter period, when temperature and relative humidity do not significantly affect survival. The results of our study provide baseline information for developing relocation guidelines for handling this species.

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